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LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW,
MX, MZ, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC,
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European patent (AT, BE, BG, CH, CY, CZ, DE, DK, EE,
ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO,
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17 June 2004

*For two-letter codes and other abbreviations, refer to the "Guid-
ance Notes on Codes and Abbreviations" appearing at the begin-
ning of each regular issue of the PCT Gazette.*

(54) Title: **USE OF GRANZYME B AS AN HSP70/HSP70 PEPTIDE DEPENDENT INDUCER OF APOPTOSIS IN TUMOR
CELLS**

(57) Abstract: The present invention relates to a method of inducing or enhancing the expression of granzyme B in natural killer (NK) cells. The present invention relates also to a use of said NK cells for the preparation of a pharmaceutical composition for the treatment of tumors, viral or bacterial infections or inflammatory diseases. Further, the present invention relates to the use of granzyme B for the treatment of tumors, viral or bacterial infections or inflammatory diseases, wherein the tumor cells or the cells affected by said infection or inflammation express Hsp70 in their cell surface.

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INTERNATIONAL SEARCH REPORT

(PCT Article 18 and Rules 43 and 44)

Applicant's or agent's file reference	FOR FURTHER ACTION see Notification of Transmittal of International Search Report (Form PCT/ISA/220) as well as, where applicable, item 5 below.	
G1437 PCT		
International application No.	International filing date (day/month/year)	(Earliest) Priority Date (day/month/year)
PCT/EP 03/09341	22/08/2003	23/08/2002
Applicant		
STEINECKE, peter		

This International Search Report has been prepared by this International Searching Authority and is transmitted to the applicant according to Article 18. A copy is being transmitted to the International Bureau.

This International Search Report consists of a total of 7 sheets.

☒ It is also accompanied by a copy of each prior art document cited in this report.

1. Basis of the report

- a. With regard to the **language**, the international search was carried out on the basis of the international application in the language in which it was filed, unless otherwise indicated under this item.

☐ the international search was carried out on the basis of a translation of the international application furnished to this Authority (Rule 23.1(b)).

- b. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international search was carried out on the basis of the sequence listing:

☐ contained in the international application in written form.

☐ filed together with the international application in computer readable form.

☒ furnished subsequently to this Authority in written form.

☒ furnished subsequently to this Authority in computer readable form.

☒ the statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

☒ the statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

2. ☒ **Certain claims were found unsearchable** (See Box I).

3. ☐ **Unity of invention is lacking** (see Box II).

4. With regard to the **title**,

☒ the text is approved as submitted by the applicant.

☐ the text has been established by this Authority to read as follows:

5. With regard to the **abstract**,

☒ the text is approved as submitted by the applicant.

☐ the text has been established, according to Rule 38.2(b), by this Authority as it appears in Box III. The applicant may, within one month from the date of mailing of this international search report, submit comments to this Authority.

6. The figure of the **drawings** to be published with the abstract is Figure No.

☐ as suggested by the applicant.

☐ because the applicant failed to suggest a figure.

☐ because this figure better characterizes the invention.

☒ None of the figures.

INTERNATIONAL SEARCH REPORT

International Application No

PCT/EP 03/09341

A. CLASSIFICATION OF SUBJECT MATTER

IPC 7 C12N5/08 A61K38/48 A61K35/14

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 C12N

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

WPI Data, PAJ, EPO-Internal, MEDLINE, BIOSIS, CHEM ABS Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 99 49881 A (MULTHOFF GABRIELE) 7 October 1999 (1999-10-07) cited in the application page 1, paragraph 1 page 3, paragraph 5 page 8, paragraph 2 -page 9, paragraph 1 page 11, paragraph 2 page 21 -page 26; examples 1-3	12,13, 16,22-24
X	US 6 261 839 B1 (MULTHOFF GABRIELE ET AL) 17 July 2001 (2001-07-17) column 1, line 65 -column 2, line 53; claims 1,19,20 -/-	12,13, 16,22-24

☒ Further documents are listed in the continuation of box C.☒ Patent family members are listed in annex.

* Special categories of cited documents :

- "A" document defining the general state of the art which is not considered to be of particular relevance
- "E" earlier document but published on or after the international filing date
- "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- "O" document referring to an oral disclosure, use, exhibition or other means
- "P" document published prior to the international filing date but later than the priority date claimed

- "T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
- "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
- "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.
- "&" document member of the same patent family

Date of the actual completion of the international search

7 April 2004

Date of mailing of the international search report

23/04/2004

Name and mailing address of the ISA

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Authorized officer

Sitch, W

INTERNATIONAL SEARCH REPORT

International Application No

PCT/EP 03/09341

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	<p>DATABASE BIOSIS 'Online! BIOSCIENCES INFORMATION SERVICE, PHILADELPHIA, PA, US; October 2001 (2001-10) MULTHOFF GABRIELE ET AL: "A 14-mer Hsp70 peptide stimulates natural killer (NK) cell activity" Database accession no. PREV200200042332 XP002276403 abstract & CELL STRESS AND CHAPERONES, vol. 6, no. 4, October 2001 (2001-10), pages 337-344, ISSN: 1355-8145</p>	
A	<p>WO 02 22656 A (MULTHOFF GABRIELE) 21 March 2002 (2002-03-21) page 1, paragraph 1 page 2, paragraph 4 -page 3, paragraph 3</p>	
X	<p>WO 99 43840 A (BOEHRINGER INGELHEIM PHARMA) 2 September 1999 (1999-09-02) page 6, line 24 -page 7, line 7 page 17, line 20 -page 18, line 14</p>	14,15, 17-24
X	<p>WO 01 80880 A (BARTH STEFAN ;ENGERT ANDREAS (DE); STOECKER MICHAEL (DE)) 1 November 2001 (2001-11-01) page 9, paragraph 4 -page 10, paragraph 1 claims 1,8,18</p>	14,15, 17-24
X	<p>SHAFFER W M ET AL: "HUMAN LYSOSOMAL CATHEPSIN G AND GRANZYME B SHARE A FUNCTIONALLY CONSERVED BROAD SPECTRUM ANTIBACTERIAL PEPTIDE" JOURNAL OF BIOLOGICAL CHEMISTRY, vol. 266, no. 1, 1991, pages 112-116, XP002276402 ISSN: 0021-9258 page 112, left-hand column, paragraph 1 -right-hand column, paragraph 3 page 115, right-hand column, paragraph 3</p>	14,15, 17-24
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INTERNATIONAL SEARCH REPORT

International Application No

PCT/EP 03/09341

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	<p>DATABASE BIOSIS 'Online! BIOSCIENCES INFORMATION SERVICE, PHILADELPHIA, PA, US; 1997 HOMMEL-BERREY GRACE A ET AL: "Granzyme B independently of perforin mediates noncytolytic intracellular inactivation of vesicular stomatitis virus" Database accession no. PREV199799800859 XP002276404 abstract & CELLULAR IMMUNOLOGY, vol. 180, no. 1, 1997, pages 1-9, ISSN: 0008-8749</p>	14,15, 17-24
P,X	<p>WO 03 007889 A (RES DEV FOUNDATION) 30 January 2003 (2003-01-30) paragraph '0015! - paragraph '0017! paragraph '0029! paragraph '0126! - paragraph '0127! paragraph '0231!</p>	14,15, 17,21,24
P,X	<p>DATABASE MEDLINE 'Online! US NATIONAL LIBRARY OF MEDICINE (NLM), BETHESDA, MD, US; MOSER ET AL: "INHIBITION OF TUMOR GROWTH IN MICE WITH SEVERE COMBINED IMMUNODEFICIENCY IS MEDIATED BY HEAT SHOCK PROTEIN 70 (HSP70) - PEPTIDE - ACTIVATED, CD94 POSITIVE NATURAL KILLER CELLS" Database accession no. NLM12653481 XP002276405 abstract & CELL STRESS AND CHAPERONES, vol. 7, no. 4, - October 2002 (2002-10) pages 365-373,</p>	12,13, 16,22
A	<p>WO 99 09206 A (BLOOD RES CENTER) 25 February 1999 (1999-02-25) page 1, line 25 -page 3, line 11 page 5, line 13 -page 6, line 33</p>	
A	<p>FR 2 792 205 A (INST NAT SANTE RECH MED) 20 October 2000 (2000-10-20) page 4, line 16 -page 5, line 31</p>	12,13
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INTERNATIONAL SEARCH REPORT

International Application No

PCT/EP 03/09341

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
T	<p>DATABASE BIOSIS 'Online! BIOSCIENCES INFORMATION SERVICE, PHILADELPHIA, PA, US; 17 October 2003 (2003-10-17) GROSS CATHARINA ET AL: "Cell surface-bound heat shock protein 70 (Hsp70) mediates perforin-independent apoptosis by specific binding and uptake of granzyme B." Database accession no. PREV200300583456 XP002276406 abstract & JOURNAL OF BIOLOGICAL CHEMISTRY, vol. 278, no. 42, 17 October 2003 (2003-10-17), pages 41173-41181, ISSN: 0021-9258</p> <p>-----</p>	

INTERNATIONAL SEARCH REPORT

International application No.
PCT/EP 03/09341

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:
Although claims 1, 2, 9-11, 21-24 (all partially), and claims 3, 16-20 (all completely) are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.
2. ☐ Claims Nos.:
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/EP 03/09341

Patent document cited in search report		Publication date	Patent family member(s)	Publication date
WO 9949881	A	07-10-1999	DE 19813760 A1 CA 2325735 A1 WO 9949881 A2 EP 1066050 A2 JP 2002509892 T	07-10-1999 07-10-1999 07-10-1999 10-01-2001 02-04-2002
US 6261839	B1	17-07-2001	DE 19813759 C1 EP 0952213 A2 JP 2000001436 A	15-07-1999 27-10-1999 07-01-2000
WO 0222656	A	21-03-2002	AU 1223602 A WO 0222656 A2 EP 1319023 A2	26-03-2002 21-03-2002 18-06-2003
WO 9943840	A	02-09-1999	AU 761497 B2 AU 2113299 A BR 9909250 A CA 2321178 A1 CN 1292032 T EP 1056877 A1 HU 0100776 A2 JP 2002504381 T NZ 506941 A PL 343248 A1 TR 200002496 T2 WO 9943840 A1 US 6537784 B1 ZA 9901519 A	05-06-2003 15-09-1999 04-09-2001 02-09-1999 18-04-2001 06-12-2000 28-06-2001 12-02-2002 30-01-2004 30-07-2001 21-11-2000 02-09-1999 25-03-2003 23-11-1999
WO 0180880	A	01-11-2001	AU 6221601 A WO 0180880 A2 EP 1283719 A2 US 2003186384 A1	07-11-2001 01-11-2001 19-02-2003 02-10-2003
WO 03007889	A	30-01-2003	WO 03007889 A2 US 2003086919 A1	30-01-2003 08-05-2003
WO 9909206	A	25-02-1999	CA 2298867 A1 EP 1007723 A1 WO 9909206 A1	25-02-1999 14-06-2000 25-02-1999
FR 2792205	A	20-10-2000	FR 2792205 A1 AT 243252 T CA 2368029 A1 DE 60003417 D1 DK 1171578 T3 EP 1171578 A2 EP 1346725 A1 WO 0063348 A2 HK 1044795 A1 JP 2003502284 T PT 1171578 T	20-10-2000 15-07-2003 26-10-2000 24-07-2003 13-10-2003 16-01-2002 24-09-2003 26-10-2000 10-10-2003 21-01-2003 28-11-2003